ACUTE TOXICITY SUMMARY

BENZENE

(benzol; benzole; cyclohexatriene)

CAS Registry Number: 71-43-2

I. Acute Toxicity Summary (for a 6-hour exposure)

Inhalation reference exposure level	**1,300 μg/m³**
Critical effect(s)	Reproductive/developmental toxicity
Hazard Index target(s)	Reproductive/developmental; Immune System; Hematologic System;

II. Physical and Chemical Properties (HSDB, 1994 except as noted)

Description	colorless liquid
Molecular formula	C_6H_6
Molecular weight	78.1
Density	0.879 g/cm^3 @ 25°C
Boiling point	80.1°C
Melting point	5.5°C
Vapor pressure	100 mm Hg @ 26.1°C
Flashpoint	-11°C
Explosive limits	upper = 8.0% by volume in air lower = 1.4% by volume in air
Solubility	soluble in ethanol, chloroform, ether, carbon disulfide, acetone, oils, and glacial acetic acid; slightly soluble in water
Odor threshold	0.875 ppm (2.8 mg/m³) (Haley, 1977)
Odor description	sweet
Metabolites	hydroquinone, quinone, catechol, phenol
Conversion factor	1 ppm = 3.24 mg/m³

III. Major Uses or Sources

Benzene has been widely used as a multipurpose organic solvent. This use is now discouraged due to its high toxicity. Present uses include benzene as a raw material in the synthesis of styrene, phenol, cyclohexane, aniline, and alkyl benzenes and in the manufacture of various plastics, resins, and detergents. Synthesis of many pesticides and pharmaceuticals also involves benzene as a chemical intermediate (HSDB, 1994). Benzene is emitted in large quantities from refineries and petroleum storage facilities. The tire industry and shoe factories use benzene extensively. Annual demand in the U.S. was estimated to be 6 million tons in 1990 (HSDB, 1994).

IV. Acute Toxicity to Humans

Deaths from acute exposure to benzene are often related to physical exertion and release of epinephrine with subsequent cardiac failure. Frequently, the person trying to rescue a collapsed victim will die during the effort of lifting the unconscious person (HSDB, 1994). Anesthesia may develop at concentrations above 3,000 ppm (9,600 mg/m³) (Reprotext, 1993). At exposures of greater than 1,000 ppm (3,200 mg/m³) (duration unspecified), CNS symptoms include giddiness, euphoria, nausea, and headaches; heightened cardiac sensitivity to epinephrine-induced arrhythmias may develop (Snyder, 1987). These effects may be accompanied by symptoms of mild irritation to the eyes and mucous membranes. Acute hemorrhagic pneumonitis is highly likely if benzene is aspirated into the lung (HSDB, 1994). Respiratory tract inflammation, pulmonary hemorrhages, renal congestion, and cerebral edema have been observed at autopsy in cases of acute benzene poisoning (IARC, 1987). In these cases, blood levels of 2 mg/ml benzene were not associated with hematological changes (Winek and Collom, 1971).

Systemic poisoning by benzene can occasionally result in neuroretinal edema and in retinal and conjunctival hemorrhage (Grant, 1986). Additionally, petechial hemorrhages of the brain, pleura, pericardium, urinary tract, mucous membranes, and skin may occur in cases of fatal, acute benzene poisoning (Haley, 1977).

Major concerns of systemic benzene toxicity include aplastic anemia and acute myelogenous leukemia (IARC, 1987; Reprotext, 1993). Both of these conditions are typically seen in the chronic and subchronic exposures, but may be of concern following acute exposures as well. Myeloid and erythroid components of the bone-marrow are specific targets of benzene toxicity, which leads to aplastic anemia (IARC, 1982).

In men and women exposed to benzene for 4 hours, 46.9% of the inhaled dose was absorbed. Of this absorbed fraction, 30.1% was retained and 16.8% was excreted unchanged in the expired air (Nomiyama and Nomiyama, 1974). Most of the catechol and phenol metabolites are excreted within 24 hours in the urine, while hydroquinone requires 48 hours (Teisinger *et al*., 1952).

Exposure at the odor threshold (0.875 ppm or 2.8 mg/m³) for a brief duration is reported to enhance the electropotential of the brain (Haley, 1977).

Predisposing Conditions for Benzene Toxicity

Medical: People with existing hematologic disorders and cellular anemias may be more sensitive to the acute toxicity of benzene to the bone-marrow (Reprotext 1993, 1999). People with heart conditions may also be at increased risk for cardiac arrhythmias induced by exposure to high levels of benzene. Administration of epinephrine is known to potentiate the cardiac toxicity of benzene (Reprotext, 1993).

Females may be more sensitive to benzene toxicity than males due to higher average body fat content, which serves as a storage reservoir for the chemical

(Reprotext, 1993). Similarly, obese individuals of either sex may be more sensitive to benzene toxicity.

Chemical: Previous acute exposure to toluene inhibits benzene metabolism to toxic metabolites, and may reduce toxicity (Reprotext, 1993). Consumption of ethanol potentiates the bone-marrow toxicity of inhaled benzene in mice (Baarson *et al*., 1982).

V. Acute Toxicity to Laboratory Animals

The oral LD_{50} in rats is reported to be 3.4 g/kg in young rats and 4.9 g/kg in older rats (Kimura *et al*., 1971). Mortality was observed in 2 out of 10 rats exposed to 33,000 mg/m³ (10,300 ppm) for 12.5-30 minutes daily for either 1 or 12 days (IARC, 1982). A 4-hour LC_{50} of 13,700 ppm (43,800 mg/m³) was reported in female rats (IARC, 1982). An LC_{Lo} of 45,000 ppm (144,000 mg/m³) is reported in rabbits (RTECS, 1994). In mice, an LC_{50} of 9,800 ppm (31,400 mg/m³) is reported (RTECS, 1994). Leukopenia has been demonstrated to occur in rabbits exposed to 240 ppm (767 mg/m³) for 10 hours/day for 2 weeks (IARC, 1982).

Brief inhalation of air saturated with benzene vapor (concentration unknown) resulted in ventricular extrasystole in cats and primates, with periods of ventricular tachycardia that occasionally terminated in ventricular fibrillation (Clayton and Clayton, 1981).

An attempt by Nielsen and Alarie (1982) to determine the inhalation RD_{50} for benzene was not successful. These investigators showed that inhalation of 5,800 ppm (18,800 mg/m³) benzene in mice caused an increase in respiratory rate beginning at 5 minutes following onset of exposure. They speculated that the stimulation of respiratory rate resulted from the action of benzene on the central nervous system. In this study, benzene was not irritating to the upper airways of the animals.

The pharmacokinetics of benzene in the rat reportedly follows a 2-compartment model. The rapid phase has an elimination half-life ($t_{1/2}$) of 0.7 hours, and the $t_{1/2}$ for the longer phase is 13.1 hours (Rickert *et al*., 1979). The long elimination half-life for benzene is due to the formation of catechol, quinone, and hydroquinone in the bone marrow. These reactive metabolites are not readily excreted, and are cytotoxic to the germinal cells in the bone marrow (Greenlee *et al*., 1981). A 3-compartment model was fitted to human data on benzene disposition and bone-marrow metabolism (Watanabe *et al*., 1994). The general relationship between cumulative quantity of metabolites produced and inhalation concentration was not linear, but was S-shaped, inflecting upward at low concentrations, and saturating at high concentrations.

Mice, particularly the DBA/2 strain, are more sensitive to myelotoxicity from benzene than are rats or rabbits (IARC, 1982). Colony-forming unit cells (CFUs; leukocyte precursors) were depleted in bone-marrow cultures taken from mice exposed to 4,610 ppm (14,950 mg/m³) benzene for 8 hours. Recovery of CFUs was noted 7 days after exposure (IARC, 1982).

In addition to myelotoxicity, acute exposure to benzene may disrupt erythropoiesis and result in genotoxicity. Erythropoiesis, as measured by uptake of radiolabeled iron in the bone-marrow, has been shown to be inhibited by subcutaneous injection of 10 mmol/kg benzene in mice (Bolcsak and Nerland, 1983).

Results from subacute exposures further illustrate the hematotoxic effects of benzene and the potential for immunotoxicity. Inhalation of 103 ppm (334 mg/m^3) benzene for 6 hours/day for 7 days by mice caused decreased spleen and marrow cellularities and decreased spleen weights (Green *et al*., 1981). Benzene inhalation at concentrations of 0, 10, 30, 100, and 300 ppm (0, 32.4, 97.3, 324, and 973 mg/m^3) for 6 hours/day for 5 days resulted in a decreased host-resistance to bacterial infection by Lysteria monocytogenes (Rosenthal and Snyder, 1985). The numbers of L. monocytogenes bacteria isolated from the spleen were increased in a dose-dependent manner on day 4 of infection. The total numbers of T- and B-lymphocytes in the spleen and the proliferative ability of the splenic lymphocytes were decreased in a dose-dependent manner by benzene exposures of 30 ppm (97.3 mg/m^3) or greater. In this study, no decrement in host resistance or immune response was observed at 10 ppm (32 mg/m^3) benzene. Later studies in mice have also shown that exposure to 10 ppm for a subacute duration does not significantly alter hematological parameters in blood, spleen, thymus, or bone marrow (Farris *et al*., 1996; 1997).

Farris *et al*. (1997) reported the hematological consequences of benzene inhalation in B6C3F1 mice exposed to 1, 5, 10, 100, and 200 ppm benzene for 6 hr/day, 5 days/week for 1, 2, 4, or 8 weeks and a recovery group. There were no significant effects on hematopoietic parameters from exposure to 10 ppm benzene or less. Thus 10 ppm was a NOAEL for 1 week of exposure (and longer). Exposure to 100 and 200 ppm benzene reduced the number of total bone marrow cells, progenitor cells, differentiating hematopoietic cells, and most blood parameters. Replication of primitive progenitor cells in the bone marrow was increased during the exposure period as a compensation for the cytotoxicity. At 200 ppm, the primitive progenitor cells maintained an increased percentage of cells in S-phase through 25 days of recovery compared with controls.

Inhalation of 3 ppm (9.6 mg/m^3) benzene for 6 hours by rats resulted in a significant increase over controls in the frequency of sister chromatid exchanges in peripheral blood lymphocytes (Erexson *et al*., 1986).

Evans *et al*. (1981) observed an increase in active behavior in the form of eating and grooming in mice following exposure to 300 ppm (960 mg/m^3) benzene for 6 hours.

VI. Reproductive or Developmental Toxicity

Coate *et al*. (1984) exposed groups of 40 female rats to 0, 1, 10, 40, and 100 ppm (0, 3.24, 32.4, 129.6, or 324 mg/m^3) benzene for 6 hours/day during days 6-15 of gestation. In this study, teratologic evaluations and fetotoxic measurements were done on the fetuses. A significant decrease was noted in the body weights of fetuses from dams exposed to 100 ppm (324 mg/m^3). No effects were observed at a concentration of 40 ppm (129.6 mg/m^3).

Keller and Snyder (1986) reported that exposure of pregnant mice to concentrations as low as 5 ppm (16 mg/m³) benzene on days 6-15 of gestation (6 hr/day) resulted in bone-marrow hematopoietic changes in the offspring that persisted into adulthood. However, the hematopoietic effects (e.g., bimodal changes in erythroid colony-forming cells) in the above study were of uncertain clinical significance. In a similar, later study, Keller and Snyder (1988) found that exposure of mice in utero to 20 ppm (64 mg/m³) benzene on days 6-15 of gestation resulted in neonatal suppression of erythropoietic precursor cells and persistent, enhanced granulopoiesis. This effect was considered significant bone-marrow toxicity by the authors. No hematotoxicity was seen in this study at 10 ppm (32 mg/m³).

An exposure of 500 ppm (1,600 mg/m³) benzene through days 6-15 of gestation was teratogenic in rats while 50 ppm (160 mg/m³) resulted in reduced fetal weights on day 20 of gestation. No fetal effects were noted at an exposure of 10 ppm (Kuna and Kapp, 1981). An earlier study by Murray *et al*. (1979) showed that inhalation of 500 ppm benzene for 7 hours/day on days 6-15 and days 6-18 of gestation in mice and rabbits, respectively, induced minor skeletal variations.

Tatrai *et al*. (1980) demonstrated decreased fetal body weights and elevated liver weights in rats exposed throughout gestation to 150 mg/m³ (47 ppm).

VII. Derivation of Acute Reference Exposure Level and Other Severity Levels (for a 1-hour exposure)

Level protective against mild adverse effects: While benzene exposure results in decreased immune response and hematopoietic effects in laboratory animals following 5 day exposures, it was problematic to extrapolate from these repeated dose studies for these endpoints. Thus, no level protective against mild adverse effects for one-hour is being recommended. The REL is based on developmental toxicity, a severe adverse effect.

Reference Exposure Level for a 6-hour exposure (Level Protective Against Severe Adverse Effects): 1,300 μg/m³

Because of the uncertainty of extrapolating from repeated exposures to a one-hour concentration, we have chosen to use a single day exposure in the reproductive studies with no time extrapolation as an REL. In the case of benzene, the REL is for a 6-hour exposure.

Study	Coate *et al*., 1984; (supported by Kuna and Kapp, 1981; Keller and Snyder, 1988)
Study population	pregnant female rats
Exposure method	inhalation of 0, 1, 10, 40, or 100 ppm
Critical effects	decreased fetal body weights
LOAEL	100 ppm
NOAEL	40 ppm
Exposure duration	6 hours per day (for 5 days)

LOAEL uncertainty factor	1
Interspecies uncertainty factor	10
Intraspecies uncertainty factor	10
Cumulative uncertainty factor	100
Reference Exposure Level	0.4 ppm (1.3 mg/m³; 1,300 μg/m³)

Pregnant female rats (40 per group) were exposed for 6 hours/day on days 6-15 of gestation to benzene concentrations of 0, 1, 10, 40, and 100 ppm (0, 3.24, 32.4, 129.6, and 324 mg/m³) (Coate *et al*., 1984). The mean fetal weights from the females treated with 100 ppm benzene were significantly decreased ($p < 0.05$) compared to controls. No teratogenic, fetotoxic, or maternally toxic effects were observed in rats exposed to 40 ppm (129.6 mg/m³) benzene or less. The 40 ppm (129.6 mg/m³) concentration is considered a NOAEL for reduced fetal weight. The value of 40 ppm for a 6-hour exposure was extrapolated to a 1-hour exposure using the equation $C^n * T = k$, where $n = 2$. The resulting 100 ppm extrapolated value was used to determine the level protective against severe adverse effects using uncertainty factors of 10 for intraspecies and 10 for interspecies variation. The level protective against severe adverse effects for benzene is therefore 1.0 ppm or 3.24 mg/m³.

Kuna and Kapp (1981) found direct teratogenic effects measured as decreased crown-rump length, exencephaly, and angulated ribs in rats when pregnant females were exposed 6 hours/day during days 6-15 of gestation to a concentration of 500 ppm. In this study, a concentration of 50 ppm during gestation resulted in lower fetal weights measured on day 20 of gestation. No fetal effects were noted at an exposure of 10 ppm (32 mg/m³). Keller and Snyder (1988) reported a NOAEL of 10 ppm for developmental hematopoietic effects in mice. The highest reported NOAEL (i.e., 40 ppm) consistent with reported LOAEL values was chosen for the derivation of the Reference Exposure Level (severe adverse effect level, in this case) for benzene.

Level Protective Against Life-threatening Effects

Svirbely *et al*. (1943) exposed mice for 7 hours to various benzene concentrations. They determined a NOAEL (0/18 animals) for lethality of 4,980 ppm and a LOAEL (3/18 animals) of 7,490 ppm. A benchmark concentration derived (BC_{05}) using a log-normal model with these data is 5,650 ppm (MLE = 6,550 ppm). A life-threatening level was calculated using these data with an uncertainty factor of 30 (10 for individual variability, and 3 for interspecies uncertainty using the BC_{05} as the starting point for the calculation). The level protective against life-threatening effects is therefore 5,650 ppm ÷ 30 = 190 ppm (620 mg/m³).

VIII. References

Baarson KA, Snyder CA, Green JD, Akumar AS, Goldstein BD, Albert RE. The hematotoxic effects of inhaled benzene on peripheral blood, bone marrow, and spleen cells are increased by ingested ethanol. Toxicol Appl Pharmacol 1982;64:393-404.

Bolcsak LE, Nerland DE. Inhibition of erythropoiesis by benzene and benzene metabolites. Toxicol Appl Pharmacol 1983;69:363-368.

Clayton GD, Clayton FE. Industrial hygiene and toxicology. 3rd ed. revised. Vol. IIB. Toxicology. New York (NY): John Wiley and Sons; 1981. p. 3260-3283.

Coate WB, Hoberman AM, Durloo RS. Inhalation teratology study of benzene in rats. In: MacFarland HN, editor. Advances in modern environmental toxicology, Vol VI. Applied toxicology of petroleum hydrocarbons. Princeton (NJ): Princeton Scientific Publishers, Inc; 1984. p. 187-198.

Cronkite EP, Drew RT, Inoue T, Hirabayashi Y, Bullis JE. Hematotoxicity and carcinogenicity of inhaled benzene. Environ Health Perspect 1989;82:97-108.

Erexson GL, Wilmer JL, Steinhagen WH, Kligerman AD. Induction of cytogenetic damage in rodents after short-term inhalation of benzene. Environ Mutagen 1986;8:29-40.

Evans HL, Dempster AM, Snyder CA. Behavioral changes in mice following benzene inhalation. Neurobehav Toxicol Teratol 1981;3:481-485.

Farris GM, Robinson SN, Gaido KW, Wong BA, Wong VA, Leonard L, Shah R. Effects of low concentrations of benzene on mouse hematopoietic cells in vivo: a preliminary report. Environ Health Perspect 1996;104(6):1275-1276.

Farris GM, Robinson SN, Gaido KW, Wong BA, Wong VA, Hahn WP, Shah R. Benzene-induced hematotoxicity and bone marrow compensation in B6C3F1 mice. Fundam Appl Toxicol 1997;36(2):119-129.

Flury F. Toxicities in modern industry: pharmacological-toxicological aspects of intoxicants in modern industry (German). Arch Exp Pathol Pharmakol 1928;138:71.

Gerarde HW. Benzene. In: Toxicology and biochemistry of aromatic hydrocarbons. Elsevier Monographs. Amsterdam, the Netherlands: Elsevier; 1960.

Grant WM. Toxicology of the eye. Springfield (IL): CC Thomas; 1986. p. 140-141.

Green JD, Snyder CA, LoBue J, Goldstein BD, Albert RE. Acute and chronic dose/response effect of benzene inhalation on the peripheral blood, bone marrow, and spleen cells of CD-1 male mice. Toxicol Appl Pharmacol 1981;59:204-214.

Greenlee WF, Sun JD, Bus JS. A proposed mechanism of benzene toxicity: formation of reactive intermediates from polyphenol metabolites. Toxicol Appl Pharmacol 1981;59:187-195.

Haley TJ. Evaluation of the health effects of benzene inhalation. Clin Toxicol 1977;11(5):531-548.

(HSDB) Hazardous Substances Data Bank. National Library of Medicine, Bethesda, MD (CD-ROM version). Denver (CO): Micromedex, Inc.; 1994. (Edition expires 1/31/94).

(IARC) International Agency for Research on Cancer. IARC monographs on the evaluation of the carcinogenic risk of chemicals to humans. Vol. 29. Some industrial chemicals and dyestuffs. Lyon: IARC; 1982. p. 93-148.

Keller KA, Snyder CA. Mice exposed in utero to low concentrations of benzene exhibit enduring changes in their colony forming hematopoietic cells. Toxicology 1986;42:171-181.

Keller KA, Snyder CA. Mice exposed in utero to 20 ppm benzene exhibit altered numbers of recognizable hematopoeitic cells up to seven weeks after exposure. Fundam Appl Toxicol 1988;10:224-232.

Kimura ET, Ebert DM, Dodge PW. Acute toxicity and limits of solvent residue for sixteen organic solvents. Toxicol Appl Pharmacol, 1971;19:699-704.

Kuna R., Kapp RW. The embryotoxic/teratogenic potential of benzene vapor in rats. Toxicol Appl Pharmacol 1981;57:1-7.

Murray FJ, John JA, Rampy L., Kuna RA, Schwetz BA. Embryotoxicity of inhaled benzene in mice and rabbits. Am Ind Hyg Assoc J 1979;40:993-998.

Nomiyama K, Nomiyama H. Respiratory elimination of organic solvents in man. Benzene, toluene, n-hexane, trichloroethylene, acetone, ethyl acetate and ethyl alcohol. Int Arch Arbeitsmed. 1974;32:85-91

Nielsen GD, Alarie Y. Sensory irritation, pulmonary irritation, and respiratory stimulation by alkyl benzene and alkylbenzenes: prediction of safe industrial exposure levels and correlation with their thermodynamic properties. Toxicol Appl Pharmacol 1982;65:459-477.

(RTECS®) Registry of Toxic Effects of Chemical Substances. National Institute of Occupational Safety and Health, Cincinnati, OH (CD-ROM version). Denver (CO): Micromedex, Inc.; 1994. (Edition expires 1/31/94).

Reprotext® System. Dabney BJ, editor. Denver (CO): Micromedex, Inc.; 1993. (Edition expires 11/31/93).

Reprotext ® System. Dabney BJ, editor. Denver (CO): Micromedex, Inc.; 1999. (Edition expires 1/31/1999).

Rickert D, Baker, TS, Bus JS, Barrow CS, Irons RD. Benzene disposition in the rat after exposure by inhalation. Toxicol Appl Pharmacol 1979;49:417-423.

Rosenthal GJ, Snyder CA. Modulation of the immune response to Listeria monocytogenes by benzene inhalation. Toxicol Appl Pharmacol 1985;80:502-510.

Snyder CA. Benzene, In: Snyder R, editor. Ethyl Browning's toxicity and metabolism of industrial solvents. Amsterdam: Elsevier; 1987. p. 3-37.

Svirbely JL, Dunn RL, von Oettingen WF. The acute toxicity of vapors of certain solvents containing appreciable amounts of benzene and toluene. J Ind Hyg Toxicol 1943;25:366-373.

Tatrai E, Ungvary GY, Hudak A, Rodics K, Lorincz M, Barcza GY. Concentration dependence of the embryotoxic effects of benzene inhalation in CFY rats. J Hyg Epidemiol Microbiol Immunol 1980;24(3):363-371.

Teisinger J, Bergerova-Fiserova V, Kudrna J. The metabolism of benzene in man (Pol). Pracov Lek 1952;4:175-188. [cited in International Agency for Research on Cancer (IARC) monographs. Vol. 29. 1987. p. 117.]

Watanabe KH, Bois FY, Daisey JM, Auslander DM, Spear RC. Benzene toxicokinetics in humans: exposure of bone marrow to metabolites. Occup Environ Med 1994;51(6):414-420.

Winek CL, Collom WD. Benzene and toluene fatalities. J Occup Med 1971;13:259-261. [cited in International Agency for Research on Cancer (IARC) monographs. Vol. 29. 1987. p. 116.]